which phenazine methosulphate is coupled to 2,6-dichlorophenol-indophenol. The pH optimum is 8.5 and the $K_m$ for trimethylamine is about 2$\mu$M. Of 42 compounds tried as substrate, the following gave an initial rate 10% or more of that with trimethylamine under the conditions tested: ethyldimethylamine, diethylmethylamine, 2-aminoethyldimethylamine, 2-hydroxyethyldimethylamine, 2-chloroethyldimethylamine and diethylamine.

The first step in the breakdown of trimethylamine by bacterium 4B6 thus appears to be the oxidative *N*-demethylation catalysed by trimethylamine dehydrogenase.

J. C. thanks the Science Research Council for a studentship.

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## The Metabolism of Trimethylamine *N*-Oxide by *Bacillus* PM6

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*Bacillus* PM6 is an aerobic spore-forming Gram-positive rod-shaped organism isolated from soil. It is capable of growth on trimethylamine *N*-oxide, trimethylamine, dimethylamine or methylamine as sole source of carbon, nitrogen and energy. Washed suspensions of trimethylamine *N*-oxide-grown organisms rapidly take up oxygen in the presence of trimethylamine *N*-oxide, dimethylamine and methylamine, but not in the presence of trimethylamine. Such suspensions, and their cell-free extracts, convert trimethylamine *N*-oxide anaerobically into dimethylamine and formaldehyde, indicating the presence of a trimethylamine *N*-oxide demethylase. Assays of trimethylamine *N*-oxide (Bystedt, Swenne & Aas, 1959), dimethylamine (by g.l.c. and paper chromatography) and formaldehyde (Nash, 1953) yield results that agree with the stoicheiometry required by the following equation:

$$(CH_3)_3NO = (CH_3)_2NH + H\cdot CHO$$

In early experiments the reaction rate was followed by measuring formaldehyde production colorimetrically (Nash, 1953); subsequently a more sensitive and convenient assay was developed in which the trimethylamine *N*-oxide demethylase is coupled to formaldehyde dehydrogenase (EC 1.2.1.1) (partially purified from baker's yeast) and the reaction is followed spectrophotometrically at 340nm.

The trimethylamine *N*-oxide demethylase was purified 175-fold. The purified enzyme moves as a single protein on analytical polyacrylamide-gel electrophoresis and has a molecular weight of 50000 as determined by gel filtration on Sephadex (Andrews, 1970). Treatment with 1% sodium dodecyl sulphate + 1% 2-mercaptoethanol caused no resolution of the protein as determined by polyacrylamide-gel electrophoresis (Weber & Osborn, 1969), suggesting that it consists of a single polypeptide chain; this experiment yielded a molecular weight of 37000. Ultracentrifugal analysis (sedimentation equilibrium) gave a molecular weight of 36000–47000. The pH optimum of the purified enzyme is 7.5, the $K_m$ for trimethylamine *N*-oxide is 2.85mM and the absorption spectrum shows only a single peak at 280nm. Ferrous iron, glutathione and L-ascorbate are strongly stimulatory.

Benzyldimethylamine *N*-oxide, chlorpromazine *N*-oxide and (+)-propoxyphene *N*-oxide were also active as substrate for the enzyme, but the maximum rates obtained were only about 5% of that obtained with trimethylamine *N*-oxide. Trimethylamine is a non-competitive inhibitor, and SKF 525-A is a competitive inhibitor.

The initial step in the metabolism of trimethylamine *N*-oxide by *Bacillus* PM6 thus appears to be a demethylation, catalysed by trimethylamine *N*-oxide demethylase, yielding dimethylamine and formaldehyde.

P.M. gratefully acknowledges the award of a Science Research Council Studentship.

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## Configurational Dependencies of [$^{19}$F]Fluorine Chemical Shifts and Coupling Constants in Fluoromonosaccharides

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In general, chemical shifts and coupling constants for $^{19}$F are of an order of magnitude greater